Investigations into the salinity preferences of successive larval developmental forms of five indigenous species of the freshwater prawn *Macrobrachium*

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Abstract

The salinity preferences of the larvae of 5 indigenous species of *Macrobrachium* namely *M. lepidactylus, M. rude, M. petersi, M. scabriculum* and *M. australe* was investigated. Results showed that saline conditions are definitely required for the survival and development of the early larval stages. Survival and morphological development of the larvae of all 5 species studied improved markedly once the salinity of the medium used was increased to levels exceeding 5 S ‰ with the most satisfactory levels being above 10 S ‰.

Introduction

The genus *Macrobrachium* Bate, 1868 occurs throughout the tropics and in several subtropical areas (Holthuis, 1980). Almost all the species spend part of their life cycle in freshwater (Holthuis, 1980) and the term "freshwater prawn" has been applied to representatives of the genus (Goodwin and Hanson, 1975). Of the 125 known species, most are of edible size and it is likely that they would be used as food wherever they occur (Holthuis, 1980).

With the exception of the proposed culture of *Macrobrachium* in Ghana (Prah, 1980), very little information appears to be available on the utilisation of freshwater prawn resources in Africa, for the purpose of aquaculture (Rabanal, 1980). Holthuis (1980) reported that the commercial culture of *M. rosenbergii* has been investigated in Malawi, Mauritius and the Seychelles.

Production of *M. rosenbergii* reached a commercial level by 1980 in Mauritius (Thompson, 1980) and by 1981 at an inland production unit for the same species in Zimbabwe (Kenmuir, 1981). Production trials with *M. rosenbergii* have also been conducted in the Transvaal (Taylor et al., 1992).

While *M. rosenbergii* (De Man) is a universal choice for commercial culture, the Directorates of Nature Conservation in South Africa are naturally concerned about the possible introduction of this prawn into South African waters where it might pose a potential threat to the indigenous species. In addition, climatic conditions over much of South Africa are not entirely satisfactory for culturing this tropical species whilst the local species may possibly provide a suitable alternative in the more temperate regions of the country. It was therefore decided to investigate aspects of factors affecting larval development of 5 of the indigenous species of *Macrobrachium*, namely:

Macrobrachium lepidactylus (Hilgendorf, 1897)
Macrobrachium rude (Heller, 1862)
Macrobrachium petersi (Hilgendorf, 1879)
Macrobrachium scabriculum (Heller, 1862)
Macrobrachium australe (Guerin, 1838).

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These species vary in their distribution, with *M. rude, M. lepidactylus* and *M. scabriculum* occurring along the east coast of Africa and Madagascar and extending as far as India. *M. australe* is distributed over a similar area (see Schoonbee et al., 1989), but extends as far as Polynesia (Holthuis, 1980). The present record of this species from the African continent (Schoonbee et al., 1989) is apparently the first according to Professor Holthuis, (Leiden, Netherlands) who identified the material. *M. petersi* appears to be restricted in distribution to the southeast coast of Africa (Holthuis, 1950; Read, 1982; 1985a,b; Coetzee, 1988; Bickerton, 1989). Available information indicates that, with the exception of *M. petersi* for which no relevant data are available, the various species of *Macrobrachium* are used for food in the areas where they occur, being fished to varying degrees (Holthuis, 1980). Fishing for *M. rude* is regular in certain parts of India and Bangladesh (Holthuis, 1980). This species is listed as a cultivated species by Panikkar (1968). Studies of the field biology of *M. rude* have been conducted (Ling and Costello, 1976), but no data on its possible commercial exploitation are available. The large-scale freshwater prawn culture became possible mainly as a result of the discovery by Ling(1962) that *M. rosenbergii* larvae required saline conditions for their survival and development. A knowledge of the salinity requirements of the various larval forms of our local *Macrobrachium* species is therefore a prerequisite for their culture.

The basis for the present series of experiments was the hypothesis of Knowlton (1974) that a hierarchy of developmental processes existed, based on the utilisation of food energy. Knowlton (1974) found that larval morphogenesis of *P. vulgaris* closely parallelled growth but not moulting history, and that there was a cessation of morphogenesis with cessation of growth. Thus evidence of morphological change was taken as an indication that the larvae were growing. For the purpose of this study it was necessary to establish whether development was taking place among the larvae being reared. As specimens were taken from communal rearing containers, no moulting history was available and it was necessary to use changes in morphology as a yardstick of development.

Materials and methods

The design of the present experiments was largely based on the

investigations conducted by Choudhury (1970b; 1971) on *M. carcinus* and *M. acanthurus* larvae.

Collection and management of adult prawns for breeding purposes

Electrofishing techniques were used for the collection of prawns from the Limpopo River system, Messina, Transvaal. The apparatus used consisted of a small portable power generator, an electrofishing unit and 2 electrodes. Captured prawns were held at the site of collection in aerated containers and were then transferred to tough nylon bags, containing water from the sources of collection. These bags were then filled with oxygen and sealed. Adult prawns, especially berried females, were packed separately at densities between 1 and 5 per 3 ℓ of water.

It took approximately 7 h to transport the prawns to the laboratory from both sampling localities. On arrival the bags were first floated on the water surface of the holding tanks for approximately 30 min in order to allow temperatures to equalise. Bags were then opened and the prawns were released.

Identification and separation of the species into their own specific tanks were based on descriptions and keys in Barnard (1950), Kensley (1972) and Holthuis (1950). Examples of the species were dispatched for positive identification to Professor LB Holthuis at the Natural History Museum, Leiden, Netherlands.

Adult prawns were maintained in freshwater, either from municipal or borehole supplies. An exception was the holding of *M. rude* stock at 3 to 14 S ‰ for a period of roughly 7 months. This species had been originally identified by us as *Macrobrachium equidens* (Dana), a brackish water species. After 6 weeks in the laboratory no berried females had appeared and the salinity was therefore raised in an attempt to stimulate reproduction. The prawns were fed on a pelleted diet containing 25% protein.

With the exception of *M. petersi* and *M. scabriculum*, adults of each species were maintained separately. Males and females of each species were stocked in communal tanks according to the numbers available, so that the usual sex ratios of 1 male to 5 females, as recommended for *M. rosenbergii*, was not possible. Smaller observation tanks were introduced later in order to monitor reproduction of the individual species more closely.

Tanks were checked daily for moult casts and berried females. Where possible, one male was stocked for every female in the observation tanks. Where this was not possible, males were introduced to females on the morning of a moult cast being observed. In some cases, where males were held together in separate tanks, the female would be transferred to the male tank for mating purposes. Berried females in communal tanks were removed to separate containers after mating once the eggs had become eyed.

Identification of larval forms

In order to follow the larval development of the 5 species selected for this study, it was considered necessary to divide the process of development into 7 morphological forms (stages) based on the appearance and development of selected features. The first 4 larval forms in the grid correspond closely to the first 4 larval stages described for *Macrobrachium* larvae in the literature (Ling, 1962; 1969a; Uno and Kwon, 1969; Choudhury 1970a, 1971; Pillai and Mohamed, 1973; Read, 1982). Morphological characters developing during later larval development have been grouped into a further 3 larval stages.

Experiments were conducted in a temperature-controlled water bath (28°C) which consisted of a glass aquarium tank measuring 182 x 45 x 45 cm equipped with a 300 W aquarium heater. Containers for the larvae were made from glass bottles inverted in the water bath (Read, 1982) and were held in position by a frame of polystyrene foam. Plastic funnels of 7 cm diameter were positioned over the openings of the containers in order to minimise evaporation. Aeration was provided to each container.

Two 50 ℓ aquarium tanks were used as stock tanks for freshwater and artificial sea water. These were each equipped with 100 W aquarium heaters and were constantly aerated. Sea water was prepared according to standard procedures. The freshwater tank contained maturated borehole water, aerated for at least 24 h.

Four salinities, each with 6 replicates were chosen, which were randomly assigned for each species and replicate. Larvae used in each experiment were all hatched from the same female of a given species. Ripe females were placed in 20 ℓ buckets containing water of the same temperature as the holding tank for spawning.

Once hatching was completed, the female was removed, and the volume of water in the bucket reduced to about 2 ℓ. Larvae were then transferred to an enamelled metal basin where they were counted out at 60 individuals per ℓ and transferred to the water bath. An aerator was placed in the basin and sea water was allowed to drip into the water, in the vicinity of the airstone. The salinity was checked at regular intervals until the lowest of the allotted salinities was reached. Addition of sea water was stopped, the aerator removed, and the required number of larvae were then counted out into bottles containing 500 mℓ of the allotted salinity. Sea water was again added and the procedure repeated until the larvae at all the salinities had been transferred to the water bath. The process of acclimation usually extended over a period of 2 to 3 h. Larvae were examined daily and dead and dying larvae were counted and removed and morphological development was checked.

The criterion selected for survival was the ability of the larvae to perform whole body movements, as opposed to movements of the heart, gills or pereiopods alone. Survivors were checked for morphological development under a dissecting microscope and then counted and returned to the containers, into which freshly mixed culture medium had been placed. Experiments were conducted for a period of 6 to 8 d, which allowed for the appearance of Form III larvae.

At each salinity level, 3 out of the 6 replicates were provided with freshly hatched *Artemia* nauplii while food was withheld from the other 3 replicates. The provision of *Artemia* nauplii was initiated on the second day of the experiment, at a rate of approximately 5 nauplii per mℓ. Although the form I larvae do not feed on the *Artemia*, references to fed and starved larvae will imply the provision of *Artemia* or withholding of *Artemia*. *Artemia* nauplii were provided daily to allotted containers, after examinination and transfer of larvae to a fresh culture medium. Temperatures were determined to within 0,5°C and salinities were corrected when necessary. Total NH_4 -N was monitored according to *Standard Methods* (1971). Standard methods were used for pH, PO_2 and PCO_2 determinations.

Experiments with *M. petersi* larvae

In each investigation the experiment was divided into 2 stages. The first was on the salinity requirements of the early larval

forms, and the second on the salinity requirements of the later larval forms.

Salinities of 0, 4, 8 and 12 S ‰ were chosen for the experiment with the early larval forms. Larvae were stocked at 20 per 500 mℓ of medium. The duration of the experiment was 8 d.

Stages (=Forms) 5 and 6 larvae, obtained from the same hatching as the first part of the experiment, but reared separately, were used in this experiment. Based on results obtained in the first part of the experiment, salinities of 8, 12, 16 and 20 S ‰ were chosen. Larvae were again stocked at 20 per 500 mℓ and were fed on *Artemia* nauplii once per day. The duration of the experiment was 8 d.

Experiments with *M. lepidactylus* larvae

The salinity preferences of the larvae of a coastal (Lake Cubhu) and inland population (Limpopo River) of *M. lepidactylus* were investigated as the possibility existed that the populations differed in their salinity preferences.

Salinities selected for the Lake Cubhu larvae were 0, 4, 8 and 12 S ‰, which covered the range from freshwater to salinities suitable for rearing *M. rosenbergii* larvae (Ling, 1969a;b). Larvae were stocked at 60 per ℓ. The duration of the experiment was 7 d.

Salinities of 0, 5, 10 and 15 S ‰ were chosen for the Limpopo River larvae of *M. lepidactylus*, as results from the previous experiment suggested that the species may have higher salinity preferences than 12 S ‰. Larvae were stocked at 60 per ℓ. The duration of the experiment was 8 d.

Experiments with *M. rude* larvae

M. rude larvae were stocked at 60 per ℓ. Salinities selected for this experiment were 5, 10, 15 and 20 S ‰. Freshwater was not included as the larvae of this species had already reared to post-larval stage in saline conditions. The duration of the experiment was 8 d.

Experiments with *M. australe* and *M. scabriculum* larvae

M. australe larvae were stocked at 60 per ℓ. Salinities of 5, 10, 15 and 20 S ‰ were chosen for this experiment. *M. scabriculum* larvae were stocked at 60 per ℓ. Positive identification of the female as *M. scabriculum* was possible as a result of information received from Prof. Holthuis on the separation of *M. petersii* and *M. scabriculum* adults. Salinities of 4, 8 and 12 S ‰ were chosen for this experiment. The investigation was terminated after 7 d.

Results

Salinity preferences of the early larval forms of *M. petersi*

Prevailing water conditions are presented in Table 1. The survival time of *M. petersi* larvae in freshwater (0 S ‰) (Figs.

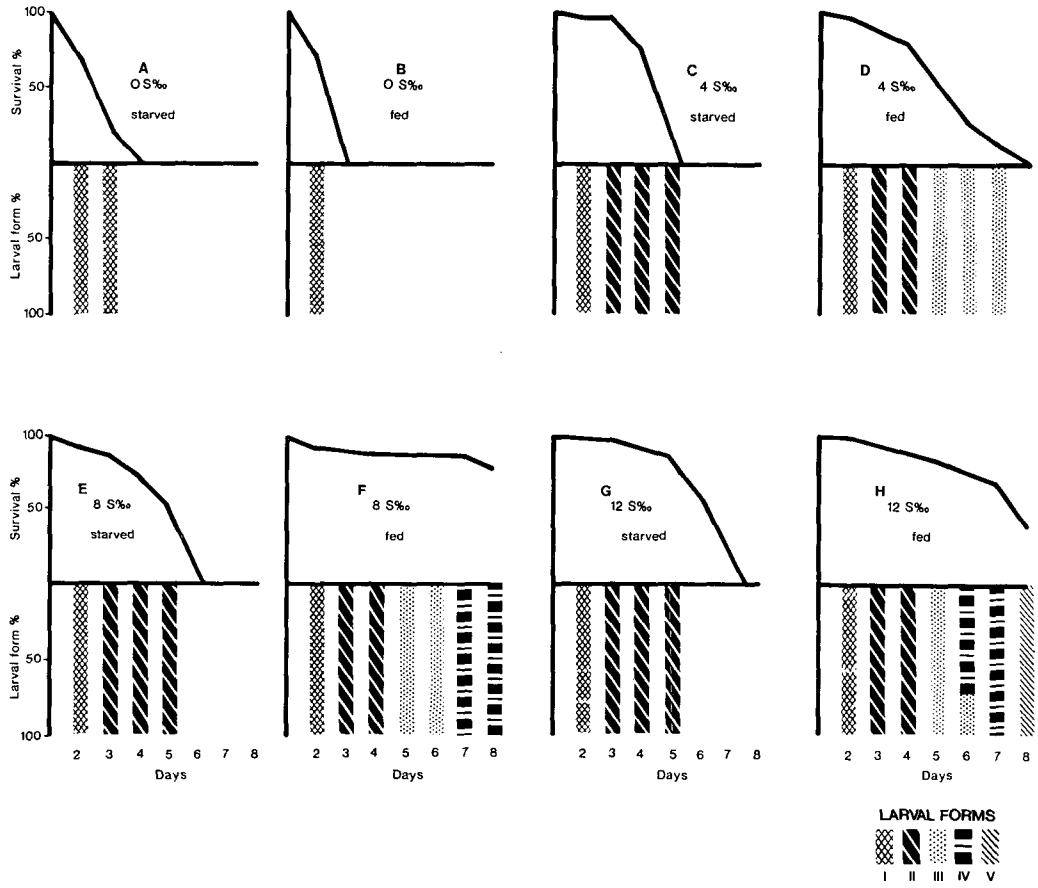

Figure 1 (A-H)
Salinity preferences of the early larval forms of Macrobrachium petersi *for starved and fed larvae at 0, 4, 8, 12 S ‰*

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TABLE 1
SUMMARY OF THE RANGE AND MEAN VALUES FOR WATER QUALITY PARAMETERS OVER THE COURSE OF THE INDIVIDUAL SALINITY PREFERENCE EXPERIMENTS

Experiment and species	Temperature Range and Mean (°C)	Range of mean daily salinities for 6 replicas at each salinity level				pH range	NH_4-N larval containers mg/ℓ	NH_4-N freshwater stock tank mg/ℓ	NH_4-N sea water stock tank mg/ℓ	pO_2 mm Hg	pO_2 mm Hg
1 *M. petersii*	27,0-31,0 29,1	0	3,8-4,0	8,0-8,0	11,7-12,0	8,0-8,5	0,036- 0,123* 0,093	–	–	116-147,5	0,5-1,5
2 *M. petersii*	26,0-30,5 28,0	7,7-9,0	11,3-12,5	15,0-16,0	19,3-20,3	7,8-8,4	0,16-1,05 0,310	–	–	125-160	0,5-0,8
3 *M. lepidactylus* (Lake Cubhu)	27,0-28,5 27,4	0	3,5-5,0	7,5-9,5	11,7-13,2	8,0-8,5	0-0,239 0,052	0-0,102 0,03	0-0,042 0,009	108-156	0,2-3,5
4 *M. lepidactylus* (Limpopo river)	27,5-29,5 27,9	0-0,3	5,0-5,3	10,0-10,0	14,3-15,2	7,9-8,6	0-0,054 0,024	0-0,01 0,005	0-0,008 0,004	–	–
5 *M. rude*	27,0-30,5 28,3	5,0-5,8	10,0-11,3	14,0-15,7	19,7-20,2	8,3-8,5	0-0,135 0,026	0-0,01 0,005	0,001- 0,12 0,007	112-142	0,5-0,9
6 *M. australe*	25,0-29,0 27,2	5,0-5,7	10,0-10,5	14,8-15,2	19,3-20,3	8,0-8,4	0,003- 0,127 0,030	0,008**	0,008**	119-135	0,8-1,0
7 *M. scabriculum*	27,5-31,0 28,7	0	4,0-4,5	8,0-8,5	11,7-12,2	–	–	–	–	–	–

*Values for fed larvae on the final day of the experiment.

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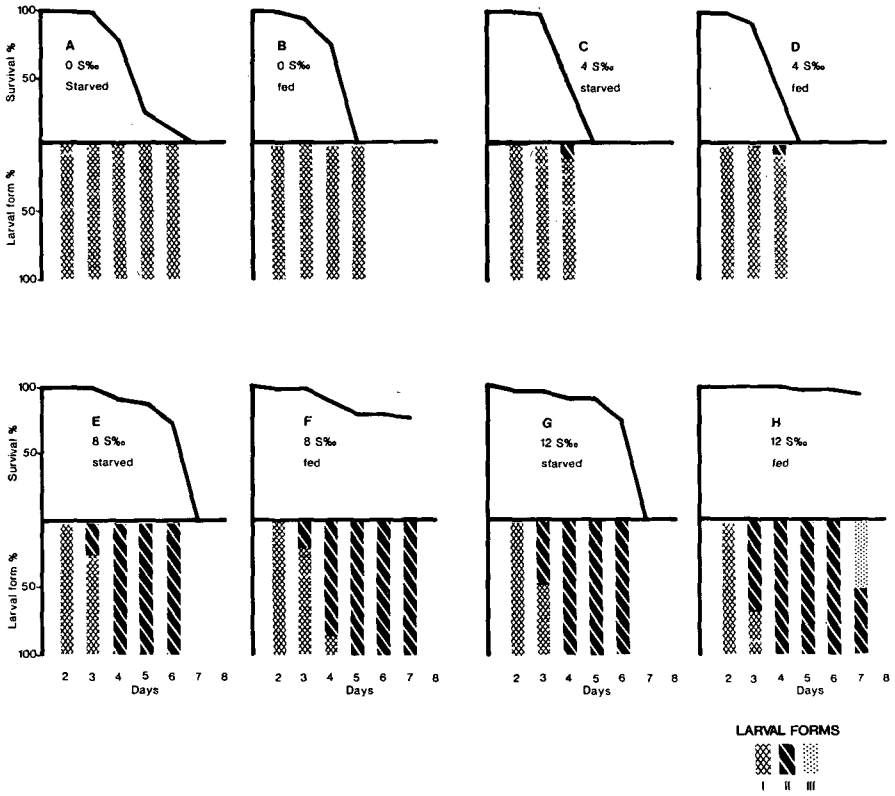

Figure 2 (A-H)
Salinity preferences of the early larval forms of Macrobrachium lepidactylus *(Coastal population, Lake Cubhu) for starved and fed larvae at 0, 4, 12 S ‰*

1A and B) was short, with 50% mortalities occurring between days 2 and 3 and 100% mortality by day 4. This applied to both fed and starved larvae, neither of which progressed beyond Form I. The survival time of starved larvae increased with increasing salinity, and a 50% mortality occurred among these larvae between days 4 and 5 at 4 S ‰, on day 5 at S ‰ and between days 6 and 7 at 12 S ‰ (Figs. 1C, E and G). Morphological development of starved larvae did not progress beyond form II at 4 S ‰ (Fig. 1C), while larvae fed *Artemia* at this salinity were able to moult to Form III, but survival decreased rapidly as the larvae reached Form III (Fig. 1D). Starved larvae at salinities 8 and 12 S ‰ were able to reach Form III but subsequently died, although survival time of Form III larvae was greater at 12 S ‰ than at 8 S ‰. Survival and morphological development of larvae was best at salinities 8 S ‰ and 12 S ‰ where the larvae were fed *Artemia* (Figs. 1F and H). Of these larvae, those held at 8 S ‰ exhibited a higher survival rate than those at 12 S ‰ (78,3% compared with 40%) on day 8, while morphological development was the most rapid at 12 S ‰ where all the surviving larvae had reached Form IV by day 8 (Figs. 1F and H).

Salinity preferences of the later larval forms of *M. petersi*

Prevailing water conditions are presented in Table 1. In all the experimental salinities, survival of larvae declined to below 50% between days 4 and 5, except for larvae in 12 S ‰. At 12 S ‰ survival approached 50% on the last day of the experiment. Although morphological development reached Form VII, no post-larvae were obtained at the termination of the experiment. This experiment was terminated as a result of the poor survival and raised NH_4-N levels (Table 1).

Salinity preferences of the early larval forms of Lake Cubhu population of *M. lepidactylus*

Prevailing water conditions are presented in Table 1. Although survival time of larvae in freshwater was slightly longer among starved larvae then larvae fed *Artemia* nauplii, 50% mortalities occurred between days 4 and 5 for both fed and starved larvae (Figs. 2A and B). Morphological development in freshwater did not progress beyond Form I.

Survival time of larvae in 4 S ‰ was shorter than in freshwater with 50% mortalities occurring among both starved and fed larvae, between days 3 and 4 (Figs. 2C and D). Morphological development of larvae at 4 S ‰ progressed to form II, but these larvae were either dead or in the process of dying when examined.

Survival time and morphological development were similar for starved larvae at 8 and 12 S ‰ respectively, with 50% mortalities occurring when the larvae had reached Form II, between days 6 and 7 (Figs. 2E and G). Morphological development did not progress beyond Form II.

The highest survival rate and the most rapid morphological development occurred where the larvae were fed *Artemia* nauplii at 12 S ‰, with a 94% survival rate by day 7. At this point approximately 50% of the survivors had reached Form III (Fig. 2H).

Salinity preferences of the early larval forms of the Limpopo population of *M. lepidactylus*

Prevailing water conditions are presented in Table 1. Survival of larvae in freshwater was similar among both fed and starved larvae, with starved larvae surviving slightly longer than larvae

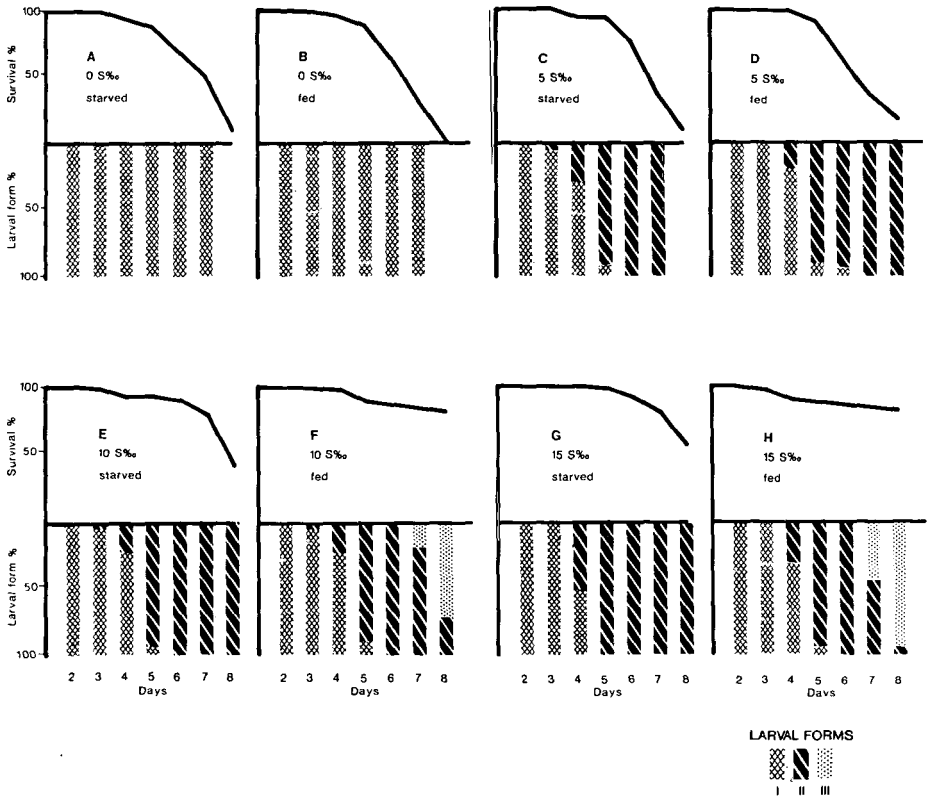

Figure 3 (A-H)
Salinity preferences of the early larval forms of Macrobrachium lepidactylus *(Inland population, Limpopo River) for starved and fed larvae at 0, 5, 10, 15 S* ‰

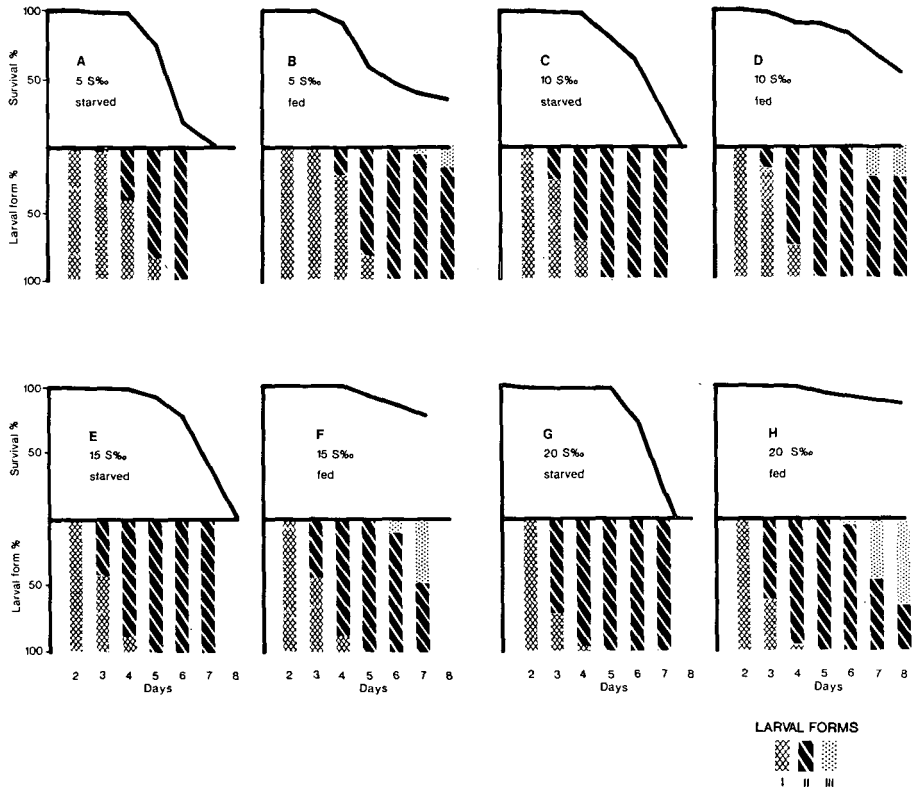

Figure 4 (A-H)
Salinity preferences of the early larval forms of Macrobrachium rude *for starved and fed larvae at 5, 10, 15 and 20 S S* ‰

provided with *Artemia* (Figs. 3A and B). The 50% mortality level was reached between days 6 and 7 where larvae were provided with *Artemia*, and on day 7 for starved larvae, but 100% mortality had not occurred in either of the freshwater treatments by day 8, the termination of the experiment. Survival at this point was 10% for starved larvae and 2,2% for fed larvae. Development in freshwater did not progress beyond Form I. Survival and development of both fed and starved larvae were similar at 5 S ‰ (Figs. 3C and D). Fifty per cent mortalities occurred between days 6 and 7 among both fed and starved larvae, when most of the larvae had progressed to Form II.

At 10 S ‰ and 15 S ‰, survival and development were similar for starved larvae, with 50% mortality occurring between days 7 and 8 at 10 S ‰, while at 15 S ‰ a survival rate of 54,9% was recorded on day 8 (Figs. 3E and G). Morphological development did not progress beyond Form II in either case.

Larvae fed *Artemia* at 10 and 15 S ‰ showed similar curves, with 82,4% and 83,4% survival by day 8 at 10 S ‰ and 15 S ‰, respectively (Figs. 3F and H). Morphological development was more rapid at 15 S ‰, with 94,7% of the larvae reaching Form III on day 8, compared with 72% present as Form III at 10 S ‰.

Salinity preferences of the early larval forms of *M. rude*

Prevailing water conditions are presented in Table 1. Survival of starved larvae at 5 S ‰ was poorer than for starved larvae at salinities of 10, 15 and 20 S ‰, with LD50 occurring between days 5 and 6 at 5 S ‰ and between days 6 and 7 at the higher salinities (Figs. 4A, C, E and G). Morphological development of starved larvae did not progress beyond Form II at any of the salinities investigated.

Survival of larvae fed *Artemia* at salinity 5 S ‰ decreased rapidly with the appearance of form II larvae, reaching 50% mortality between days 5 and 6, as was the case with starved larvae at this salinity (Figs. 4A and B). Survival of larvae fed *Artemia* at salinities 10, 15 and 20 S ‰ did not decline below 50% by the termination of the experiment at 8 d (Figs. 4D, F and H). Survival rate of the latter larvae was highest at 20 S ‰ where 85% survival was recorded at the termination of the experiment.

Morphological development of all fed larvae (Figs. 4B, D, F and H) progressed to Form III, but with increasing proportions of Form III larvae with increasing salinity. At 20 S ‰, 66,7% of surviving larvae fed *Artemia* had progressed to Form III, compared with 15,6% at 5 S ‰.

Salinity preferences of the early larval forms of *M. australe*

Prevailing water conditions are presented in Table 1. Survival of starved larvae was similar at all salinities with 50% mortalities occurring between days 4 and 5 (Figs. 5A, C, E and G), once all the survivors had reached Form II. When 100% mortalities are considered for starved larvae, however, survival time is longest at salinities 10, 15 and 20 S ‰.

Survival of larvae fed *Artemia* was similar at 10, 15 and 20 S ‰ but poorest at S ‰, with 50% mortality occurring between days 5 and 6 (Figs. 5B, D, F and H). In addition, morphological development at 5 S ‰ did not proceed beyond Form II. Survival of fed larvae at 10, 15 and 20 S ‰ was between 80 and 90% by day 6. However, differences in morphological development were evident, with the percentages of Form III larvae present at the termination of the experiment being lowest at 10 S ‰ (1,3%) and highest at 20 S ‰ (29,6%).

Salinity preferences of the early larval forms of *M. scabriculum*

Prevailing water conditions are presented in Table 1. Survival time of starved larvae was slightly longer than fed larvae in freshwater, with 50% mortalities occurring between days 5 and 6 for starved larvae and between days 4 and 5 for larvae provided with *Artemia* (Figs. 6A and B). Morphological development did not progress beyond Form I in freshwater.

Larvae fed *Artemia* at 4 S ‰ survived longer than starved larvae with 50% mortalities occurring between days 5 and 6 for starved larvae and between days 6 and 7 for fed larvae (Figs. 6C and D). Morphological development did not progress beyond Form II in either starved or fed larvae.

Survival and development of starved larvae were similar at 8 and 12 S ‰, with 50% mortality occurring between days 6 and 7 at 8 S ‰ and on day 7 at 12 S ‰ (Figs. 6E and G). Morphological development did not progress beyond Form II and was more rapid at 12 S ‰ than at 8 S ‰.

Survival of larvae fed *Artemia* at 8 and 12 S ‰ was similar, with 90,1% and 86,7% survival at 8 S ‰ and 12 S ‰ respectively at the termination of the trial (Figs. 6F and H). Morphological development was faster at 12 S ‰, where 48,7% of the surviving larvae reached Form III at the termination of the experiment, compared with 14,6% at 8 S ‰.

Discussion

In the studies of the effects of salinity and/or temperature on survival and development of palaemonid larvae under laboratory conditions, NH_4-N, pO_2 and pCO_2 levels are usually not monitored, particularly if water is changed daily (Choudhury, 1970b, 1971; Sandifer, 1973; Knowlton, 1974; Dugan et al., 1975; Guest and Durocher, 1979). Choudhury (1970b) did not monitor pH in the experimental rearing of *M. carcinus*, where water was changed daily.

Dugan et al. (1975) transferred larvae daily to clean containers, when determining optimal growth and survival conditions for survival and development of the early larval forms. Larvae held at low salinities, or in freshwater, exhibited poor survival and limited morphological development at the prevailing water temperatures in the experiments.

The morphological development of the larvae of all 5 species did not progress beyond the first larval form in freshwater. Although the first larval forms of *Macrobrachium* species survive on the contents of the yolk sac and do not ingest food from the outside, *Artemia* nauplii were provided to the larvae in freshwater. This was done so that food would be available should any of the larvae progress beyond the first larval form in freshwater. This, however, had a possible negative effect, as the *Artemia* were unable to survive in freshwater and their decomposition may have negatively influenced the survival times of larvae held in freshwater and provided with *Artemia* nauplii.

The survival time of *M. petersi* larvae in freshwater, at a mean temperature of 29,1°C, was within the range obtained by Read (1982; 1986) for *M. petersi*, that is 3,4 d median survival time at 29,7°C in 0,05 S ‰. The survival time of the larvae of the two *M. lepidactylus* populations in freshwater is of interest. Larvae of the Limpopo River population exhibited a distinctly longer survival time in freshwater than the coastal population from Lake Cubhu. Numbers of larvae of the Lake Cubhu population had

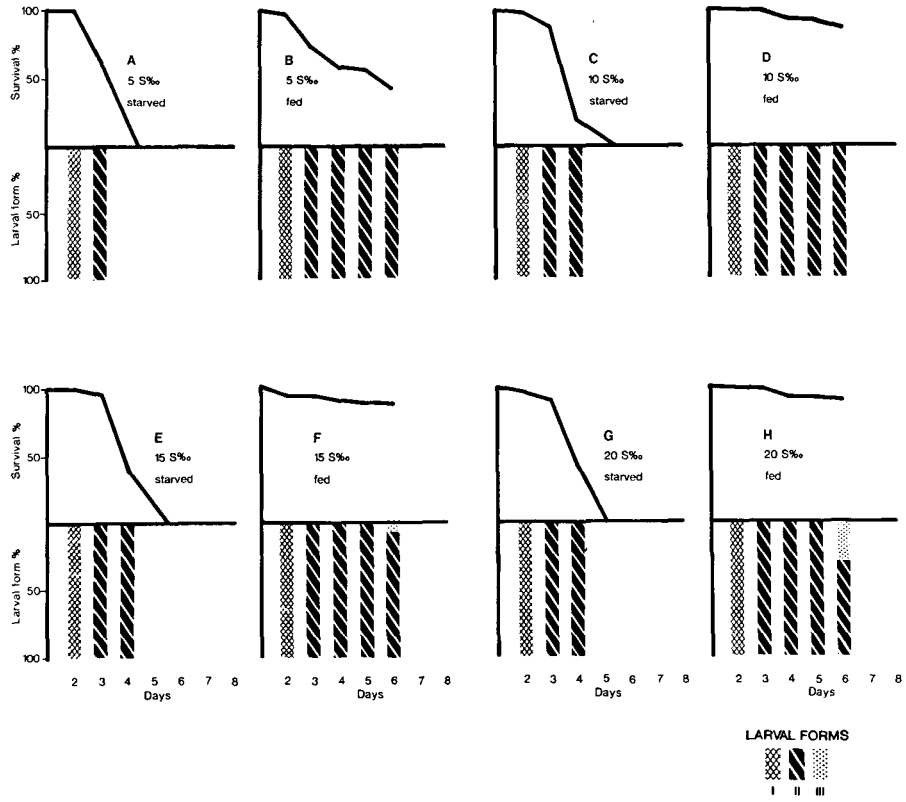

Figure 5 (A-H)
Salinity preferences of the early larval forms of Macrobrachium australe *for starved and fed larvae at 5, 10, 15 and 20 S ‰*

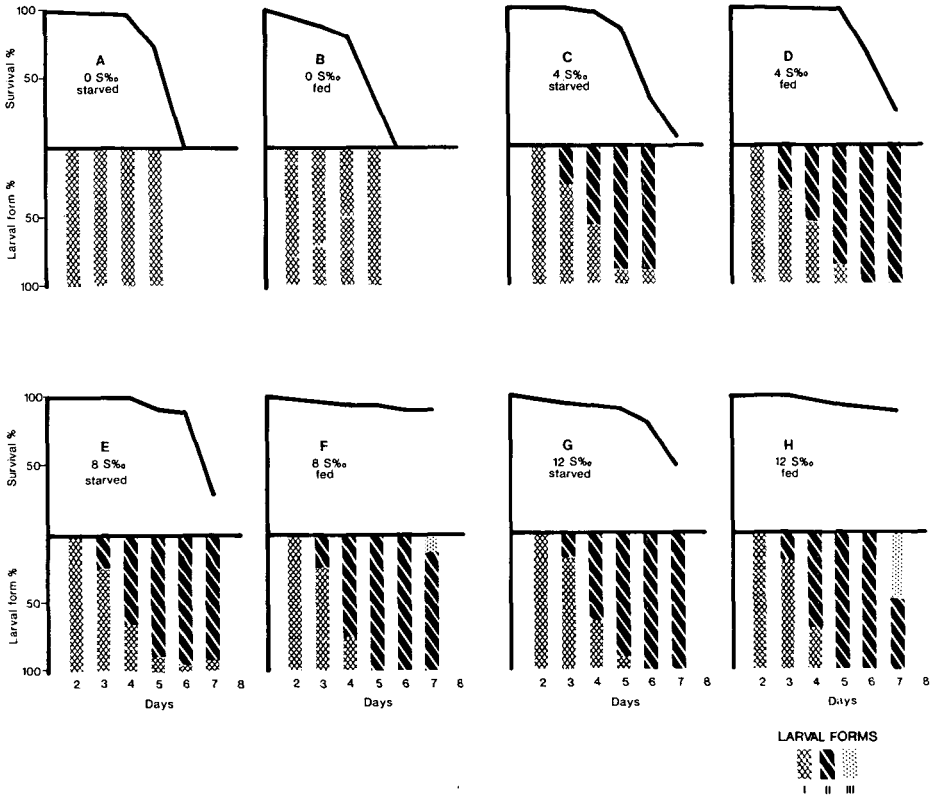

Figure 6 (A-H)
Salinity preferences of the early larval forms of Macrobrachium scabriculum *for fed and starved larvae at 0, 4, 8, 12 S ‰*

dropped to 50% between the 4th and 5th days of the experiment, while survival of the Limpopo River population was still close to 90% for the same period. None of the larvae from the Lake Cubhu population survived beyond the 6th day while survival of the Limpopo River population larvae was between 60 and 70% on the 6th day of the experiment. Since adults of the Limpopo River population were collected several hundred kilometers from the coast, and since the larvae require saline conditions for development, it seems likely that the ability to survive for a longer period of time in freshwater would be important for survival, until the larvae reached brackish water conditions. It is uncertain at this stage whether larvae are transported during flood periods to the coast, or whether the adults are catadromous, migrating down the Limpopo River to saline conditions for breeding, as is the case for *M. rosenbergii*.

M. scabriculum larvae exhibited a similar survival time to the larvae of the *M. lepidactylus* population from Lake Cubhu, that is a 50% mortality between days 5 and 6.

Survival of the larvae of all 5 species was poorest at low salinities between 4 and 5 S ‰, even where food was provided. Although larvae were able to moult to the second larval form at these salinities, survival of the second larval form was not favoured by the low salinity. *M. petersi* larvae were able to reach Form III at 4 S ‰ where *Artemia* were provided but were unable to survive. This is in agreement with the results obtained by Read (1982; 1986) which showed that low salinity and high temperature do not favour survival of *M. petersi* larvae. *M. lepidactylus* larvae from the Lake Cubhu population moulted to the second larval form at 4 S ‰ but died very soon after moulting.

Read (1982; 1986) found that salinities from 8 to 35 S ‰ were favourable for the survival of *M. petersii* larvae and that a minimum of 8 S ‰ was required for complete larval development. The results obtained for *M. petersi* larvae at 8 S ‰ are in agreement with this finding, but the poorer survival of larvae of this species at 12 S ‰ appears to be contradictory. Morphological development was favoured at the 12 S ‰ salinity level, however, with larvae reaching Form V by the 8th day. The sudden increase in mortalities with the appearance of Form V larvae may have been a result of the unsuitable nature of the experimental system for later larval forms. This was the case for Form V and VI larvae of *M. petersi*, where survival was found to be poor.

Salinities below 8 S ‰ did not favour survival of *M. lepidactylus* larvae from the Lake Cubhu population and *M. scabriculum* larvae. Salinities below 10 S ‰ did not favour survival of *M. lepidactylus* larvae from the Limpopo River population nor *M. rude* and *M. australe* larvae. It seems reasonable to assume that salinities in the region of 8 to 10 S ‰ are close to the lower limits for survival of larvae of all 5 indigenous species at temperatures of 28±1°C.

In all the experiments, morphological development was most rapid for larvae fed *Artemia* and held at the highest salinity level in each experiment. As both survival and development of larvae was favoured by the highest salinity levels in the experiments, it may be concluded that the optimum salinity levels for survival and development may in fact exceed the levels chosen for the experiments. The range of salinities tested in the apparatus used was limited by the size of the experimental set-up and should ideally include salinities from freshwater to full strength sea water. Thus further investigation of the salinity preferences of the 5 indigenous species is required. In addition, the effects of temperature in combination with salinity should be investigated, so that optimum temperature and salinity combinations for survival and development can be established for all the stages of larval development.

The poor survival of larvae of the indigenous species, during the first phase of this investigation, is attributed mainly to the low salinity levels chosen for rearing. With the exception of *M. rude* larvae, salinity levels chosen for rearing were below 10 S ‰, and often below 8 S ‰. The results from the salinity preference experiments demonstrated that higher salinity levels were required than those employed. It was nevertheless possible to rear *M. rosenbergii* and *M. rude* larvae to post-larval form, during this first phase, using biofiltration systems. This indicated that the rearing techniques employed, which included a diet of an egg custard mix and *Artemia* nauplii, would be a satisfactory base for the further rearing of larvae, once the results of the salinity preference studies became available.

A dramatic change in the survival and morphological development of *M. petersi* larvae resulted from increasing the rearing salinity to levels above 8 S ‰, considered the minimum required salinity for development of this species by Read (1982). This may be related to the osmoregulatory capacity of the larvae of *M. petersi* (Read, 1984) linked to prevailing salinities in the estuaries where it occurs along the East Coast of Southern Africa (Read 1985a;b). As in results obtained by Read for the latter species, a relatively wide range of salinities was suitable for metamorphosis to post-larvae. *M. petersi* larvae were reared to post-larval form at mean salinites ranging from 8,3 to 14,1 S ‰, at temperatures of approximately 27°C and mean salinities of approximately 10 S ‰. *M. petersi* larvae completed development in 21 d in several batches which reached post-larval form. This compares favourably with results obtained by Read (1982; 1986) for this species, namely 20 to 22 d at 25 ± 1,2 °C and salinity range 14±2,2 S ‰.

Minimum levels of 8 to 10 S ‰ were necessary for the survival and development of the other 4 species, as the results of the salinity experiments demonstrated. However, the salinity levels chosen for rearing these species, on the basis of the preferences of the early larval forms, did not produce dramatic changes in survival and development as with *M. petersi*. Read (1982; 1986) demonstrated that metamorphosis to post-larvae could take place over a wide salinity range for *M. petersi* larvae, and that the range was probably wider than for most *Macrobrachium* species. It is quite likely that the salinity requirements of the other 4 indigenous species investigated in the present study may be more restricted in range, as is the case with certain other *Macrobrachium* species. Further investigations into the salinity preferences of the later larval forms of *M. lepidactylus, M. rude, M. scabriculum* and *M. australe* are required. This should include the investigation of the effect of different salinity temperature combinations.

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